

FULL COMPATIBILITY AND FERTILITY OF *POLYPORUS* *ARCULARIUS* FROM SPAIN AND COSTA RICA.

Maria NUNEZ

Department of Biology, Division of Botany, University of Oslo, P.O.
Box 1045, Blindern, N-0316 Oslo 3, Norway.

ABSTRACT - Monospore isolates of *Polyporus arcularius* Batsch:Fr. from Spain and Costa Rica were paired and found to be genetically compatible. Basidiocarp formation following compatible mating was achieved in one isolate. The basidiospore progeny was viable and responded to the expected sexual polarity of the species. The taxonomic circumscription of *P. arcularius* based on macro- and micromorphological characters is consistent with the results obtained from compatibility tests.

RÉSUMÉ - Les mycéliums monospermes de *Polyporus arcularius* Batsch: Fr. d'Espagne et de Costa Rica ont été confrontés et se sont révélés être compatibles. La formation d'un basidiocarpe a été obtenue après confrontation. La descendance des basidiospores est viable et répond à la polarité typique de l'espèce. La délimitation taxonomique de *P. arcularius* basée sur des caractères macro- et micromorphologiques est confirmée par les résultats des tests de compatibilité.

INTRODUCTION

P. arcularius Batsch:Fr. is a cosmopolitan species with variable morphological features (Gilbertson & Ryvarden, 1987). The species is heterothallic and tetrapolar, and has heterocytic nuclear behavior (David & Romagnesi, 1972).

Basidiocarp production in culture from sympatric populations has been reported (Kitamoto et al., 1968). However, the role of geographical isolation as a source of genetic or morphological divergence in this species has never been evaluated.

MATERIALS AND METHODS

Basidiocarps of *P. arcularius* were collected from two localities:

- Bosque de la Hoja, Heredia Province, Costa Rica. 19 June 1991. MN63.
- Road from Miraflores to Puerto de La Morcuera. Madrid. Spain. 6 June 1991. MN68.

Voucher specimens are deposited in the Herbarium of Oslo University (O).

A spore suspension from sporeprints of each basidiocarp was spread over 9 cm Petri dishes with Water Agar (KEBO Lab AB).

Germings were picked under a binocular microscope and transferred to 2% Malt Extract Agar (MEA).

Three monospore isolates each from Costa Rica and Spain, chosen at random were paired at room temperature on 2% MEA (Table I). Polarity tests were not performed with them beforehand.

ISOLATES	63.1	63.2	63.3
68.1	+	+	+
68.2	+	+	+
68.3	+	+	+

Table 1. - Pairings between monospore isolates from Spain (68) and Costa Rica (63).

Tableau 1. - Confrontation des mycéliums monospermes d'Espagne (68) et de Costa Rica (63).

Two weeks after contact between monospore mycelia had been established, pairings were assessed for the presence of clamps. Mycelian nuclei were coloured following the procedure described by Laane (1970).

Secondary mycelium from the interface zone of each pairing was subcultured to confirm the constancy of clamps.

To achieve pilear development from one of the positive matings where a stipe started to form, ventilation and humidity were controlled following Kitamoto et al. (1968).

A polarity test was performed with nine monospore isolates from the basidiocarp developed in culture to confirm the tetrapolar heterothallism previously reported for this species (Vandendries, 1923).

RESULTS

Clamps and binucleate mycelium were consistently observed in all pairings between monospore isolates from Costa Rica and Spain (Fig. 1). A stable, secondary mycelium began to spread radially in the Petri dish (Fig. 2).

One week after the 63.2 x 68.1 secondary mycelium covered the Petri dish, a stipe was initiated and a pileus was formed within five days (Fig. 3).

The polarity test performed with basidiospores from this basidiocarp gave the results shown in Table 2. Occurrence of either common A or common B incompatibility factors (Takemaru, 1961) was not detected.

A comparison between several morphological characters of the basidiocarps from Costa Rica, Spain and the 63.2 x 68.1 isolate respectively shows that the radial arrangement of pores and spore size are the only consistent characters (Table 3).

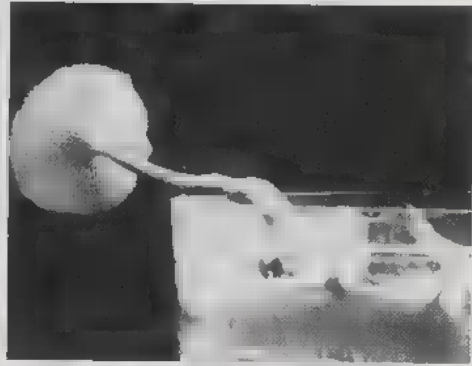
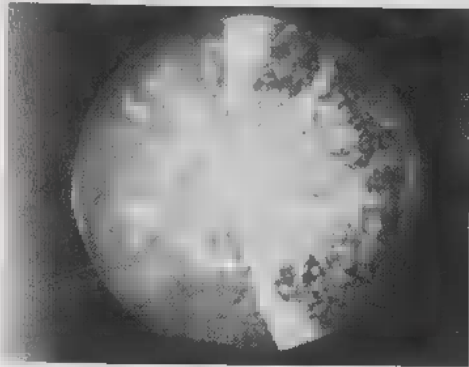


Figure 1. Hyphae from the secondary mycelium formed after compatible pairing of isolates from Spain and Costa Rica (63.2 x 68.1). Two nuclei per cell, and clamp connections can be observed.

Figure 1. - Hyphes du mycélium polysperme obtenues après la croissance compatible des cultures d'Espagne et de Costa Rica (63.2-68.1). On peut observer des hyphes binucléées et des boucles.

Figure 2. - Secondary mycelium formed after 63.2 x 68.1. The mycelial growth proceeds stably, and no somatic incompatibility zone has been formed.

Figure 2. - Mycélium polysperme obtenu après 63.2 x 68.1. Le mycélium se développe uniformément. Aucune surface d'incompatibilité n'est formée.

Figure 3. Basidiocarp developed from the secondary mycelium shown in Fig. 2. The basidiocarp was fertile (see text).

Figure 3. - Basidiocarpe obtenu à partir du mycélium polysperme de la Figure 2. Le basidiocarpe est fertile.

	1ss	6ss	7ss	8ss	2ss	4ss	3ss	5ss	9ss
1ss		-	+	+	-	-	-	-	-
6ss			+	+	-	-	-	-	-
7ss				-	-	-	-	-	-
8ss					-	-	-	-	-
2ss						-	+	+	+
4ss							+	+	+
3ss								-	-
5ss									-

Table 2. Polarity tests between monospore isolates from the basidiocarp obtained in culture. "Single spore isolate" is abbreviated as "ss". A - sign indicates incompatibility.

Tableau 2. - Tests de polarité entre des mycéliums monospermes du basidiocarpe obtenu en culture. "Culture monosporme" a été abrégée comme "ss". Le signe - indique l'incompatibilité.

ISOLATE	PILEUS COLOUR	PILEUS	MARGIN	RADIAL PORES	HYPHAL PEGS	PORE SIZE	SPORE SIZE (μ m)
MN63	dark brown	glabrous	ciliate	+	+	1-2 per mm	7-9 \times 2.5
MN68	yellow	villose	nude	+	-	1-2 per mm	7-9 \times 2-2.5
CULTURE	yellow	glabrous	nude	+	-	>1 per mm	7-9 \times 2.5

Table 3. Comparison of morphological features between collections of *P. arcularius* from Costa Rica (MN63), Spain (MN68) and the one produced in culture. A + sign indicates presence of ■ feature. A - sign indicates absence.

Tableau 3. - Comparaison des caractères morphologiques entre des récoltes de *P. arcularius* de Costa Rica (MN63), d'Espagne (MN68) et de celle obtenue en culture. Le signe + indique la présence du caractère. Le signe - indique l'absence du caractère.

DISCUSSION

The concept of "biological species" in fungi (Vandendries, 1923) is widely accepted as the basis of species delimitation (Boidin & Lanquetin, 1977; Hallenberg, 1984). Mating compatibility is considered sufficient to indicate cospecificity, due to the difficulty of producing basidiocarps in culture (see opposite cites).

Ainsworth et al. (1992) provided evidence of genetic heterogeneity in the secondary mycelia of outcrossing populations of *Stereum*, a case not found in *P. arcularius*.

Compatibility tests are useful in delimiting the distribution area of a species on a wide basis. As in the case of *P. arcularius*, geographic isolation may not be the factor that initiates the speciation process, even if gene flow between allopatric populations is substantially nonexistent (Levin, 1979).

The observed morphological differences may be due to phenotypic adaptations to environmental factors, or to genetic differences that do not interfere with mating processes (Vilgalys, 1991).

Production of basidiocarps and viable basidiospores after positive mating of *P. arcularius* demonstrates that compatibility in this species also comprises fertility.

ACKNOWLEDGEMENTS

J. Santos is acknowledged for providing material from Spain, C. Whist for taking the photograph in Fig. 3, and L. Ryvarden, A. Masuka and A.D.M. Rayner for comments that improved this manuscript. The Norwegian Agency for Science and Humanities and the Spanish Ministry of Foreign Affairs provided financial support.

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